

that is programmed into it. Alternatively, for instance, the ground can be mounted on a frame that moves in the x, y, and z planes with respect to a stationary micropipette tip streaming collagen.

5 The substrate onto which the materials are streamed, sprayed or sputtered can be the grounded target itself or it can be placed between the micropipette tip and the grounded target. The substrate can be specifically shaped, for instance in the shape of a nerve guide, skin patch, fascial sheath, or a vascular graft for subsequent use in vivo. The electroprocessed compositions can be shaped to fit a defect or site to be filled. Examples include a site from which a tumor has been removed, an injury site in the skin (a cut, a biopsy site, a hole or other defect) and a missing or shattered piece of bone. The electroprocessed compositions may be shaped into shapes useful for substance delivery, for example, a skin patch, a lozenge for ingestion, an intraperitoneal implant, a subdermal implant, the interior lining of a stent, a cardiovascular valve, a tendon, a ligament a dental prosthesis, a muscle implant, or a nerve guide. Electroprocessing allows great flexibility and allows for customizing the construct to virtually any shape needed. Many matrices are sufficiently flexible to allow them to be formed to virtually any shape. In shaping matrices, portions of the matrix may be sealed to one another by, for example, heat sealing, chemical sealing, and application of mechanical pressure or a combination thereof. An example of heat sealing is the use of crosslinking techniques discussed herein to form crosslinking between two portions of the matrix. Sealing may also be used to close an opening in a shaped matrix. Suturing may also be used to attach portions of matrices to one another or to close an opening in a matrix. It has been observed that inclusion of synthetic polymers enhances the ability of matrices to be heat sealed.

Other variations of electroprocessing, particularly electrospinning and electroaerosoling include, but are not limited to the following:

1. Using different solutions to produce two or more different fibers or droplets simultaneously (fiber or droplet array). In this case, the single component solutions can be maintained in separate reservoirs.
2. Using mixed solutions (for example, materials along with substances such as cells, growth factors, or both) in the same reservoir(s) to produce fibers or droplets composed of electroprocessed materials as well as one or more substances (fiber composition "blends"). Nonbiological but biologically compatible material can be mixed with a biological molecule.

3. Utilizing multiple potentials applied for the different solutions or the same solutions.

4. Providing two or more geometrically different grounded targets (i.e. small and large mesh screens).

5 5. Placing the mold or mandrel or other ungrounded target in front of the grounded target.

6. Applying agents such as Teflon onto the target to facilitate the removal of electroprocessed materials from the target (i.e. make the material more slippery so that the electroprocessed materials do not stick to the target).

10 7. Forming an electroprocessed material that includes materials applied using multiple electroprocessing methods. For example, electrospun fibers and electroaerosol droplets in the same composition can be beneficial for some applications depending on the particular structure desired. This combination of fibers and droplets can be obtained by using the same
15 micropipette and solution and varying the electrical charge; varying the distance from the grounded substrate; varying the polymer concentration in the reservoir; using multiple micropipettes, some for streaming fibers and others for streaming droplets; or any other variations to the method envisioned by those of skill in this art. The fibers and droplets can be layered or mixed together in same layers. In
20 applications involving multiple micropipettes, the micropipettes can be disposed in the same or different directions and distances with reference to the target.

8. Using multiple targets.

All these variations can be done separately or in combination to produce a wide variety of electroprocessed materials and substances.

25 The various properties of the electroprocessed materials can be adjusted in accordance with the needs and specifications of the cells to be suspended and grown within them. The porosity, for instance, can be varied in accordance with the method of making the electroprocessed materials or matrix. Electroprocessing a particular matrix, for instance, can be varied by fiber
30 (droplet) size and density. If the cells to be grown in the matrix require a great deal of nutrient flow and waste expulsion, then a loose matrix can be created. On the other hand, if the tissue to be made requires a very dense environment, then a dense matrix can be designed. Porosity can be manipulated by mixing salts or other extractable agents. Removing the salt will leave holes of defined sizes in
35 the matrix.

One embodiment for appropriate conditions for electroprocessing fibrin is presented below. For electroprocessing fibrin by combining fibrinogen and thrombin, the appropriate approximate ranges are: voltage 0-30,000 volts; pH 7.0 to 7.4; calcium 3 to 10 mM; temperature 20 to 40°C; ionic strength 0.12 to 0.20 M; thrombin 0.1 to 1.0 units per ml; and fibrinogen 5 to 25 mg/ml. For electroprocessing fibrin monomer, the pH starts at 5 and increases to 7.4 while the ionic strength starts above 0.3 M and decreases to 0.1 M. The other conditions are similar as stated within this paragraph. Electroprocessed fibrin matrices of varying properties can be engineered by shifting the pH, changing the ionic strength, altering the calcium concentration, or adding additional polymeric substrates or cationic materials. For electroprocessing collagen, the appropriate approximate ranges are: voltage 0-30,000 volts; pH 7.0 to 8.0; temperature 20 to 42°C; and collagen 0 to 5 mg/ml. Electroprocessed collagen matrices of varying properties can be engineered by shifting the pH, changing the ionic strength (e.g. addition of organic salts), or adding additional polymeric substrates or cationic materials.

Shapes of Electroprocessed Materials and Matrices

Electroprocessed materials can be electrodeposited inside a specifically shaped mold. For instance, a particular type of organ or tissue that to be replaced has a specific shape, such as a skin patch to fit a biopsy site or a large scalp area following a wide area removed after discovering a malignant melanoma. That shape is then reproduced and created inside a mold designed to mimic that shape. This mold can be filled by electrodepositing the material into it. In this way, the matrix exactly mimics the mold shape. In some embodiments, matrices that will become extracellular matrices and that have a specific shape are used in the creation of a new organ. Hollow and solid organs can be made. Mixing cells with the material during electrospraying forms cells within the matrix so that they do not have to migrate into a matrix.

Methods of Combining Substances with Electroprocessed Materials

Substances can be combined with the electroprocessed materials by a variety of means. In some embodiments, the substance comprises molecules to be released from the electroprocessed material and is therefore added to or incorporated within the matrix of electroprocessed material. Substances can be